

Date: 6/4/14 People in lab: Caleb Treccazi

Title: Isolation of colonies from May chemical transformations

Start Time: 9 pm

Purpose: Prep for blue-white screening of hmp

Protocol: LTM Ed. 2 Inoculation

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
I 1 6/4	Isolation of colonies for blue white screening	CT 1 5/29	1
I 2 6/4	Isolation of colonies for blue white screening	CT 2 5/29	1
I 3 6/4	Isolation of colonies for blue white screening	CT 3 5/29	1
I 4 6/4	Isolation of colonies for blue white screening	CT 4 5/29	1
I 5 6/4	Isolation of colonies for blue white screening	CT 5 5/29	1
I 6 6/4	Isolation of colonies for blue white screening	CT 6 5/29	1

Results: None of the isolations had expected growth

Notes:

Stop Time: 10 pm

Next: PCR of hmp again

Date: 6/6/14 People in lab: Caleb Treccazi

Title: Digest of chloramphenical resistant vector

Start Time: 7 pm

Purpose: To have the correct backbone ready for parts when done

Protocol:LTM Ed. 2 Digests

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
Chlor Vector 6/6	Chlor resistant backbone cut at EcoRI and pstI	iGEM Distribution Kit 2012	1

Results:

Notes:

Stop Time: 9 pm

Next: Ligation of norV A1 and A2 with chlor vector 6/6

Date: 6/6/14 People in lab: Caleb Treccazi

Title: Ligation of norV into chlor resistant backbone

Start Time: 9 pm

Purpose: To have a functional part

Protocol: LTM Ed. 2 Ligation

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
L 1 6/6	norV gene in chlor vector	A 1 12/2 and chlor vector 6/6	1
L 2 6/6	norV gene in chlor vector	A2 12/2 and chlor vector 6/6	1

Results:

Notes:

Stop Time: 9:15 pm

Next: Chemical transformations

Date: 6/7/14 People in lab: Emily Puleo, Kira Buckowing, Caleb Treccazi

Title: PCR of hmp gene

Start Time: 10:30 am

Purpose: To add the iGEM prefix and suffix

Protocol: LTM Ed. 2 PCR

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
6/7 hmp A	---	---	1
6/7 hmp B	---	---	1

Results:

Notes:

Stop Time: 11 am

Next: Gel electrophoresis

Date: 6/7/14 People in lab: Emily Puleo, Kira Buckowing, Caleb Treccazi

Title: Gel electrophoresis of hmp PCR products

Start Time: 11:30 am

Purpose: To check for products

Protocol: LTM Ed. 2 Gel

Exceptions:

Products:

Well	1	2	3	4	5	6	7	8
-	-	6/7 hmp A	-	Ladder	-	6/7 hmp B	-	

Products:

Results:

Notes:

Stop Time: 12: 15 pm

Next: TOPO

Date: 6/7/14 People in lab: Emily Puleo, Kira Buckowing, Caleb Treccazi

Title: TOPO cloning of hmp

Start Time: 11:45 am

Purpose: To make more copies of amplified hmp

Protocol: LTM Ed. 2 Chemical Transformations and Invitrogen TOPO cloning kit manual

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
6/7 CT hmp A1-1	20 uL	6/7 hmp A	1
6/7 CT hmp A1-2	200 uL	6/7 hmp A	1
6/7 CT hmp A2-1	20 uL	6/7 hmp A	1
6/7 CT hmp A2-2	200 uL	6/7 hmp A	1
CT hmp B1-1	20 uL	6/7 hmp B	1
CT hmp B1-2	200 uL	6/7 hmp B	1
CT hmp B2-1	20 uL	6/7 hmp B	1
CT hmp B2-2	200 uL	6/7 hmp B	1

Results: (6/8) No growth seen - cells not competent or died?

Notes:

Stop Time: 3:30 pm

Next: Make competent cells (ran out); try PCR once more

Date: 6/8/14 People in lab: Caleb Treccazi

Title: Chemical Transformation of 6/6 L1 and L2

Start Time: 10:30 am

Purpose: To check for successful product from ligation

Protocol: LTM Ed. 2 Chemical Transformation

Exceptions:

Products:

Sample Label	Description	Source Label	Quantity
CT 1 6/8 norV A1 and chlor R	norV gene with chlor resistant backbone	L 1 6/6	1
CT 2 6/8 norV A2 and chlor R	norV gene with chlor resistant backbone	L 2 6/6	1
CT 3 negative control	Dead cells	Competent cells and chlor plate	1
CT 4 6/8 positive control	Live cells	Comp cells and amp plate	1

Results: no growth besides on the positive control plate where there were very few colonies

Notes: Old comp cells were used that were found in the back of the -80 C freezer; cell competency is suspect and experiment needs repeating with new competent cells and new plates

Stop Time: 1:30 pm

Next: Repetition with new materials